

Effect of salinity and food ration level on the growth of Nile tilapia (*Oreochromis Niloticus* L.)

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Abstract

The effect of salinity (0, 10 & 20‰, water temperature $28 \pm 1^\circ\text{C}$) and food ration (3 and 4.5% bw/day) on food consumption and growth of Nile tilapia, *Oreochromis niloticus* ($10.77 \pm 0.21\text{g}$) were investigated. Individual food consumption was measured using X-radiography technique. Salinities (0, 10 & 20‰) did not have significant effect on the growth rate of groups of Nile tilapia fed at different ration levels (3 & 4.5% bw/day). This study showed that the growth of all-male fish was significantly better than all-female fish for all three salinities and two rations. Salinities from 0 to 20‰ had no effect on growth performance of males or female fish. In the present study, it was evident that fish fed at 3% bw/day ration ate all the food offered and fish fed at 4.5% bw/day did not consumed all amounts. Also, growth performance did not significantly differ among fish fed at 3% bw/day ration level and reared at different salinities. Fish reared under higher salinities (20‰) and fed at higher ration (4.5% bw/day) level had skin lesions and injuries on their body. It was assumed that fish fed at higher ration under higher salinities (20‰) and maintained higher osmoregulatory costs together with osmotic stress may have a negative influence on the appetite of fish. Another possibility that may have affected the appetite could be the unionized ammonia levels that were high. The high-unionized ammonia levels combined with the osmotic stress may have been the cause, or have aided, development of skin lesions and injuries on the fish at higher salinities.

Keywords: Nile tilapia, Salinity, Ration, X-radiography technique, Growth

Introduction

Ration level is one of the most important determinants of fish growth. In the absence of dietary input, no growth occurs so ration level is necessary to satisfy the energy requirements of food processing and basal metabolic activity. The first rule of fish feeding is do not overfeed (Stickney 1986). In general, overfeeding is practiced as a strategy in the early life stages of many cultivated species including tilapias but should be avoided during on-growing not only because of the cost of wasted feed but because of the potential environmental impact of that waste. From the aquaculture point of view, however, it is essential to optimise ration levels for particular species to maximize an efficient and cost-effective production cycle.

In recent years, with the increased intensification of culture methods for tilapia species it has become necessary to provide ration levels of food that satisfy their nutrient requirements for all stages of development and for both freshwater and brackishwater. Numerous studies have been conducted in freshwater to help determine optimum ration levels for tilapia species. Coche (1982) found that Nile tilapia sized 5-20 g needs 14-12% bw/day ration and fish sized 20-40 g need 6.0-4.5% bw/day ration at temperature 28°C but temperatures of 5°C or below these feeding rates should be decreased in line with growth rate and appetite. Clark *et al.* (1990) indicated that Florida red tilapia feeding rates decline from about 10.5% bw/day for 10 g fish through 7.5% bw/day for 30 g fish, 5.0 % bw/day for 60 g fish, 4.5% bw/day for 90 g fish, to around 3.5% bw/day for 150 g fish.

Ali (2001) indicated that fish reared in salinities up to 20‰ did not show significant differences of food consumption and growth performance of *O. niloticus* when feeding at 2% bw/day ration level and also suggested that 2% bw/day ration was a restricted level for juvenile *O. niloticus*. Although some experiments have been conducted to determine the effects of ration levels of *O. niloticus* under freshwater culture systems, to date there has been no attempt to investigate this particular species under controlled conditions using it in combination with salinity and ration levels for individuals reared in a group. It is, however, evident that the effect of salinity on growth will be difficult to determine without specific knowledge of food consumption. X-radiography (Ali 2001, Stead *et al.* 1999) allows investigators to examine food consumption rates of individual fish reared in larger groups. The aim of this study is to investigate the effect of ration level and salinity on the relationships between food consumption rates, growth rates and feed conversion ratios. In addition, the differences in growth between male and female fish under different salinities and rations were examined.

Materials and methods

Experimental set up

The experiment was conducted between 11 March and 10 May 2001, at the Department of Zoology, University of Aberdeen, Scotland, UK. Three treatments were designated as T₁ (freshwater 0‰, 3% ration and 4.5% ration), T₂ (10‰, 3% ration and 4.5%) and T₃ (20‰, 3% ration and 4.5%). The stocking density was maintained at the rate of 20 fish per 60L tank. Nile tilapia (*Oreochromis niloticus*) fingerlings of the same age and size were obtained from Stirling University Aquaculture Unit, Stirling, Scotland, UK. The lengths (8.37±0.08 cm) and weights (10.77 ±0.21 g) of each individual fish were measured before randomly dividing the fish into twelve 60L experimental tanks, so that there were 20 fish in each tank. Each individual fish was freeze branded (marked) by liquid nitrogen (Ali 2001). Each fish could be individually identified by using different combinations of freeze branded marks for easy monitoring. The three different treatments of salinities and food rations (3 and 4.5% bw day⁻¹ once a day in the morning between 9:00-9:30h) were all duplicated. Tank water was passed through a box-

type gravel clinoptilolite external water filter. The water temperature was maintained between 27 and 29°C, and the photoperiod was regulated by a time clock adjusted so that there was 12 h light and 12 h darkness.

After stocking, fish fed at two ration levels (3 and 4.5% bw/day) for the respective tanks were allowed to acclimate for a period of 18 days. On following day, the second measurement of lengths and weight of fish (lengths 2 and weights 2) were recorded and fish were remarked (if necessary) and replaced to their respective tanks. After the second sampling day (day 19) salinity was increased for tanks T₂ and T₃ by adding crude salt at the rate of 2‰/day until the required salinity was reached. Salinity of T₁ and T₂ was reached within 5 and 10 days respectively and on days 29 the lengths and weights were measured again and the fish returned to their respective tanks for individual food consumption rates to be measured.

Diet and feeding regime

Normally the fish were hand-fed a commercial pellet (45% protein, oil 18%, ash 8.5%, fiber 2%; "Ewos" fish food company, UK). The mean body weight of all fish in each tank was used to calculate the ration level given daily to the fish. For the measurement of individual food consumption, the normal feed was replaced by feed containing radio-opaque ballotini (size 30, 0.40-0.60mm, British Optical Ltd. Wal sall). The marked feed was prepared by grinding the normal feed, and ballotini (2.5% of the food wet weight) and water (15% of the food wet weight) were added. The feed was then mixed for three hours and repelleted as normal feed and dried overnight at 70°C and stored at 0°C until required. To calculate the relationship between the amount of food consumed by each fish and the number of ballotini present in the digestive tract of each fish, a calibration line was calculated for the marked feed before the experiment commenced. This was carried out by X-raying known weights of food (0.025-2.701 g) and counting the number of ballotini (X-ray negative) contained. A regression line was then constructed relating the number of ballotini to a weight of food ($Y = 0.0138X - 0.039$, $n = 38$, $R^2 = 0.99$; $p < 0.05$). The diet calibration curve was constructed and the high correlation of the regression line suggested the uniformity of the labeling of the feed with ballotini.

Measurements of individual food consumption

Individual food consumption was measured on three sampling occasions (Day 40, 50 & 61), using the X-radiography technique (Ali 2001, Stead *et al.* 1999). On the day that consumption rates were determined, the ballotini labeled diet (3 and 4.5% in the respective tanks) used for food consumption measurements. One hour after feeding, the fish were anaesthetised, identified by marked reading, X-rayed and weighed to the nearest 0.1 g. X-ray films were then developed and the amounts of feed consumed estimated from the counts of the number of ballotini present in the gastrointestinal tracts of each individual fish. Following completion of feed intake, lengths and weights measurements, the fish were returned to their respective tanks. In order to determine the

“Accountability” of food eaten versus not eaten by the fish, as determined by X-ray measurements the wet weight of food eaten by all fish (as shown on the X-ray negatives) in each tank was calculated and then divided by the wet weight of food offered in each tank and the results were expressed as percentages (%).

Sex identification

At the end of the experiment, the sex (male and female) of each fish was identified by observation using the hand/manual sexing procedure of Balarin and Hatton (1979). This technique is used because the structure of the genital papillae is indicative of sex. When weighing more than 20 g, the sex of this species is easily identifiable by their genital papillae. In males, there is a single genital opening on the tip of the papillae where as in females the genital opening is separated and is located on the frontal wall of the papilla, which is close to the apex.

Treatments of data and statistical analyses

The specific growth rates, SGR for individual fish were calculated by using the equation of Ricker (1979). The FCR was calculated as described by Stead *et al.* (1999). Food consumption, FC for individual fish was determined using the following equation: FC (mg dry weight of food) = GB* X; where, GB is the number of glass beads observed in each fish based on one X-ray measurement and X is the milligrams of dry food corresponding to one glass bead from the calibration curve. Condition factors (K) were determined according to the formula: $K (g\ cm^{-3}) = W / L^3 * 100$; where, W is wet weight of fish (g) and L is the final fork length of fish (cm). Two-way analysis of variance (ANOVA) was used to examine the differences in weights, lengths, specific growth rates of male and female fish, food consumption rates and feed conversion ratios within (ration levels) and between (salinity) treatments. Data are presented as means together with their standard errors.

Results

Growth performance of Nile tilapia *O. niloticus* was observed during the experimental period from 3 months. No significant differences ($p > 0.05$) were found in initial lengths (cm) and body weights (g) of fish in treatments for the two rations (3 and 4.5% bw/day) or within the different salinities (0, 10 and 20‰) (Table 1). Mean final lengths (cm) and weights (g), SGR and condition factors were (Table 1) not significantly different ($p > 0.05$) between replicate tanks for the two rations or within different salinities. Since there were no significant differences of weight changes in replicate tanks then the average was taken to produce the graphs (Figs. 1a & 1b) for the fish reared in different salinities and fed at two ration levels. There were no significant differences ($p > 0.05$) in weight gain observed for fish under different salinities and in fish fed on different ration levels of 3% bw/day (Fig. 1a) and 4.5% bw/day (Fig. 1b). The fish fed at 3% bw/day ration level showed that as the salinity increased the rate of weight increases comparatively reduced (Fig. 1a). Fish fed at 4.5% bw/day ration level showed no

significant differences in weight increase for 0 and 20‰ salinity. However, the fish in 10‰ salinity seemed to increase slight faster in weight although there were no significant differences ($p > 0.05$) were observed.

To explain the observed trends of growth rates, the experimental period has been divided into 2 halves (29 & 32 days for 1st and 2nd half, respectively). Figs. 2a & 2b showed that SGR calculated using consecutive sampling days (i.e. SGR-1 calculated using W_0 and W_1 and time was the number of days between weighing). For both ration levels (3 and 4.5% bw/day) SGR reduced in the second half of the experiment. Fish fed at 3% bw/day ration level and reared in freshwater (0‰) seem to maintain a higher SGR in the second half of the experiment than fish reared in 10 and 20‰. Fish fed at 4.5% bw/day ration level showed that fish reared in 10‰ seem to have a higher SGR in the first half of the experiment.

An individual rate of food consumption was based on each X-ray measurement. Accountability (%) of the X-ray measurements was very low. Table 2 showed that the food consumed on each of the three sampling days was low for all tanks irrespective of the salinity or food ration given. Feed conversion ratio was also low with fish showed lower SGR. Values for feed conversion ratios were expected to be higher for lower specific growth rates. The water quality of all tanks in the present study was similar which means the experimental conditions were similar. Unionized ammonia (mg/L) levels were always found to be above 0.80 mg/L (data not shown).

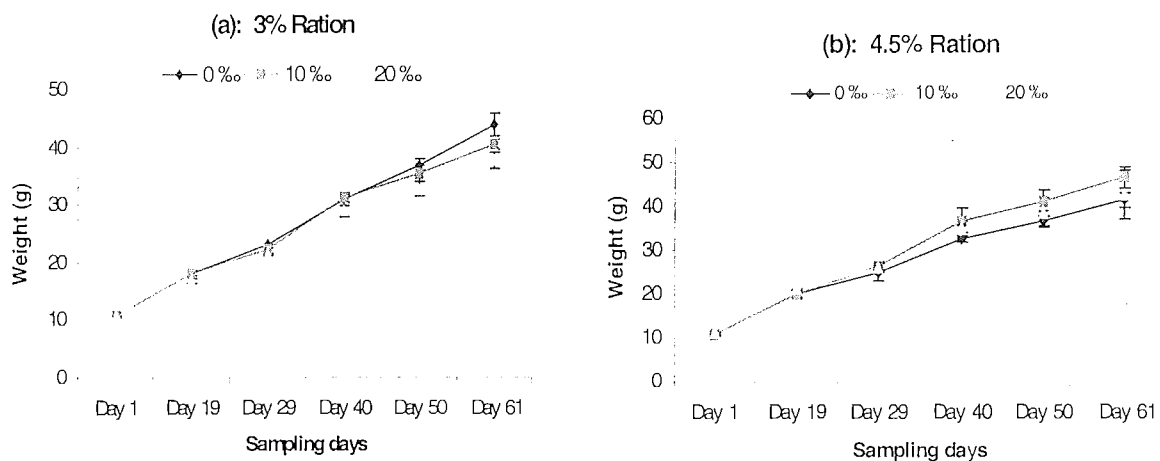


Fig. 1. The increase of wet weight of *O. niloticus* in different salinities (0, 10 & 20‰) at 3% bw/day ration in (a) and 4.5% bw/day ration in (b) during the experimental period.

Table 1. Summary of the growth performance of *O. niloticus* during the experimental period. Mean initial and final lengths and weights, mean specific growth rates and mean condition factors calculated for all individual fish for each tank (n = 20) and shown as means of two tanks under each salinity and ration. Mean values sharing common superscripts (down columns) are not statistically different at the 5% significance level (Duncan's multiple comparison test)

Treat ment	Salinity (%)	Ration (% bw/day)	Initial length (cm)	Final length (cm)	Initial weight (g)	Final weight (g)	SGR _m (%/day)	CF _m (g/cm ³)
T ₁	0	3	8.40 ± 0.07 ^a	12.87 ± 0.32 ^a	10.74 ± 0.24 ^a	43.87 ± 1.42 ^a	2.20 ± 0.08 ^a	1.92 ± 0.02 ^a
		4.5	8.41 ± 0.08 ^a	12.73 ± 0.28 ^a	10.75 ± 0.26 ^a	41.32 ± 1.69 ^a	2.02 ± 0.02 ^a	1.93 ± 0.02 ^a
T ₂	10	3	8.28 ± 0.07 ^a	12.74 ± 0.26 ^a	10.81 ± 0.29 ^a	40.45 ± 0.56 ^a	2.04 ± 0.08 ^a	1.91 ± 0.03 ^a
		4.5	8.38 ± 0.07 ^a	13.28 ± 0.29 ^a	10.77 ± 0.28 ^a	46.55 ± 3.97 ^a	2.28 ± 0.15 ^a	1.90 ± 0.02 ^a
T ₃	20	3	8.31 ± 0.08 ^a	12.48 ± 0.21 ^a	10.58 ± 0.24 ^a	37.65 ± 0.96 ^a	2.09 ± 0.16 ^a	1.89 ± 0.03 ^a
		4.5	8.41 ± 0.08 ^a	12.95 ± 0.28 ^a	10.95 ± 0.27 ^a	42.60 ± 1.14 ^a	2.12 ± 0.08 ^a	1.93 ± 0.04 ^a

Table 2. The rates of food consumption, accountability of X-ray measurements and feed conversion ratio of *O. niloticus* reared in different salinities (0, 10 & 20‰) and ration (3 and 4.5% bw/day) levels during the experimental period

Treatments		FC (mg /g/day)			Accountability (%)			FCR (mg/g)		
Salinity (%)	Ration (% bw/day)	FC-1	FC-2	FC-3	X-ray 1	X-ray 2	X-ray 3	FCR-1	FCR-2	FCR-3
0	3	6.40	6.44	4.46	30.30	27.22	18.92	9.89	18.88	8.42
	4.5	2.48	4.96	2.88	8.28	12.91	7.90	3.72	14.46	7.47
10	3	4.17	2.10	2.29	21.10	8.52	9.61	5.06	4.98	5.69
	4.5	5.04	2.95	1.75	16.53	7.91	4.64	2.80	3.69	1.55
20	3	3.58	1.98	1.19	16.55	8.15	3.66	5.81	7.45	3.32
	4.5	5.43	2.10	1.48	16.38	4.25	3.06	8.98	7.13	5.01

Table 3. Summary of the sex ratio of male and female, mean initial and final weight and mean specific growth rate of male and female fish under different salinities (0, 10 & 20‰) and ration (3 and 4.5% bw/day) levels during the experimental period. Mean values sharing a common superscript (down columns) are not statistically different at the 5% significance level (Duncan's multiple comparison test).

Salinity (‰)	Ration (% bw/day)	Sex ratio (%)		Mean initial weight (g)		Mean final weight (g)		SGR _m (%/day)	
		Male	Female	Male	Female	Male	Female	Male	Female
0	3	48	52	11.03 ± 0.78 ^a	10.37 ± 0.72 ^a	52.92 ± 3.15 ^a	32.46 ± 2.33 ^b	2.57 ± 0.16 ^a	2.00 ± 0.09 ^b
10	3	49	51	10.54 ± 0.54 ^a	11.04 ± 0.97 ^a	51.94 ± 4.52 ^a	35.77 ± 2.75 ^b	2.66 ± 0.34 ^a	1.96 ± 0.13 ^b
20	3	47	53	10.91 ± 0.82 ^a	10.29 ± 0.50 ^a	47.05 ± 2.98 ^a	32.83 ± 1.99 ^b	2.44 ± 0.12 ^a	1.93 ± 0.09 ^b
0	4.5	51	49	11.08 ± 0.71 ^a	10.43 ± 0.67 ^a	50.54 ± 3.87 ^a	33.06 ± 2.37 ^b	2.53 ± 0.31 ^a	1.92 ± 0.24 ^b
10	4.5	47	53	11.88 ± 1.12 ^a	10.71 ± 0.98 ^a	52.7 ± 4.97 ^a	35.41 ± 1.25 ^b	2.63 ± 0.21 ^a	1.99 ± 0.12 ^b
20	4.5	48	52	11.09 ± 0.99 ^a	10.79 ± 0.36 ^a	48.48 ± 2.68 ^a	33.92 ± 2.27 ^b	2.46 ± 0.22 ^a	1.91 ± 0.25 ^b

Effect of salinity and food ration level on the growth of Nile tilapia

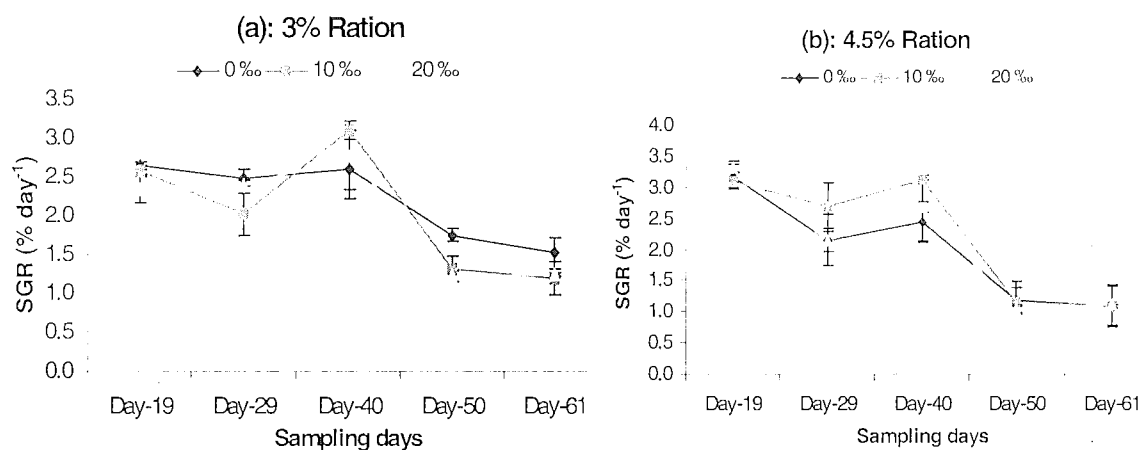


Fig. 2. Influence of salinity on specific growth rates at 3% and 4.5% bw/day ration levels over the experimental period of 61 days. SGR of fish is calculated for individuals and then presented as a mean SGR for 3% bw/day ration level (mean of replicates tanks where $n = 40$ fish) are taken to produce the graphs in (a) and similarly 4.5% bw/day ration level in (b).

At the end of the experiment, sex was identified by hand and data are shown in Table 3. Males and females ratio (%) and initial weights (g) of males and females were more or less similar in all treatments and replicated tanks. Male fish grew faster than females in case of all salinities and ration levels. Final weight gain (g) and specific growth rates (SGR, %/day) of males were significantly ($p < 0.05$) higher than females within the same salinity (0, 10 & 20‰) and within the same ration levels (3 and 4.5% bw/day) but there were no significant differences between ($p > 0.05$) the all-male fish in different salinities and ration levels and all-female fish in different salinities and ration levels.

Discussion

Tilapias have been known to feed up to 7% of their own body weight (Jauncey 1998). Since specific growth rate (SGR, %/day) seems to stabilise at 4 to 5% bw/day ration level (Shouqi *et al.* 1997) the 3% and 4.5% bw/day ration levels were chosen to ensure good growth rates. During the first half (29 days) of the experiment, the fish consumed all or most of the ration, however in the second half (32 days) of the experiment the percentage of eaten reduced. Those fed at 3% bw/day ration level usually had less trouble in consuming the full amount, but 4.5% bw/day ration level was usually too much and the water quality may have deteriorated where ammonia levels were above 0.80 mg/L.

Many of the tanks had fish attaining, or had already attained sexual maturity towards the end of the first half of the experiment. Therefore, when or if there were any females carrying eggs in their mouths they will not eat. During the experiment and on

sampling days some fish in the tanks were found with eggs or young fry. Few eggs or young fry being found does not determine the amount of eggs actually produced because eggs could have been eaten by the mother (due to poor water quality or if eggs are not fertilised) or any of the other fish (cannibalism) (Bolivar *et al.* 1993). So females carrying eggs or young in their mouth may have influenced the amount of food eaten by the group as a whole. It was observed that male and female ratios were similar in all tanks. Balarin and Hatton (1979) reviewed the characteristics and environmental requirements of some important tilapia species (including *O. niloticus*) used in fish culture and cited that in the wild, male fish grows 1-5 times faster than female fish. In the present study, significantly ($p < 0.05$) higher growth rates of all-male fish compared to all-female fish were observed for fish reared in different salinities and at both ration levels.

Fish reared under higher salinities (20‰) and fed a higher ration (4.5% bw/day) level had skin lesions and injuries on their body. To maintain higher osmoregulatory costs together with osmotic stress may have a negative influence on the appetite of fish (Suresh and Lin 1992). Another possibility that may have affected the appetite could be the unionised ammonia levels that were high. The high-unionised ammonia levels combined with the osmotic stress may have been the cause, or have aided, development of for the skin lesions and injuries found on the fish (Prunet and Bornancin 1989) at higher salinities (20‰). Nile tilapias have been reported to thrive in low salinities, i.e. brackishwater (Stickney 1986). According to Shouqi *et al.*, (1997) 3% bw/day ration level is just under optimum for achieving high rates of specific growth. The 4.5% bw/day ration level is just past this point. This would mean that at 4.5% bw/day ration level extra food is available to invest energy in other areas where it may be needed without affecting growth too much. One example could be when sexual maturity is attained; extra energy is needed for gonadal development. Normally, growth rates decrease when tilapia reaches sexual maturity (diverting energy to gonadal development rather than to somatic growth) (Boliver *et al.* 1993).

In the present study, food consumption and feed conversion ratios were very low probably due to low accountability of the X-ray measurements. The accountability was low for the repelleted food containing the ballotini beads. One of the reasons that could explain this is palatability of the repelleted diets. The fish were fed the same diet throughout the experiment, but only on the days that the fish were going to be X-rayed were the fish fed with pellets that contained the ballotini beads. As explained in some guides' e.g., McCarthy *et al.* (1993) for experiments involving radiography the diet should be prepared in the same way ensuring an equal physical texture for the duration of the experiment.

Shouqi *et al.* (1997) found that increased ration (0.5, 1, 2, 4 % bw/day and satiation) levels resulted in food consumption efficiencies increasing first, and then decreasing at higher ration levels. In the present study, no such consistent correlation was observed between the fish fed with 3% and 4.5% ration levels under different salinity treatments. Shouqi *et al.* (1997) also observed that Nile tilapia fed to satiation allocated 76% metabolic energy to HE (Heat Energy), and 24% to RE (Recover Energy) and suggested that this species has low growth efficiencies with high metabolic expenditure with

higher food rations. In addition, Meyer-Burgdorff *et al.* (1989) suggested that food efficiencies decreased with increasing ration level. It may be assumed from the present study that 4.5% bw/day for one meal is too much for *O. niloticus* and that some fish did not actually eat this entire amount. As a result, no significant differences in growth were observed between fish on higher (4.5% bw/day) & lower (3% bw/day) ration levels.

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